6-2-2001

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Euphytica **119:** 95–100, 2001.

Breeding for end-use quality: Reflections on the Nebraska experience

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Key words: Genotypic and phenotypic selection, glutenin subunits, wheat breeding

Abstract

Every cultivar released in Nebraska must have four characteristics: improved agronomic performance relative to existing cultivars, exceptional winterhardiness, resistance to *Puccinia graminis* (the causal agent of stem rust), and acceptable end-use quality. This paper will discuss our strategy for breeding cultivars with acceptable end-use quality. All experimental lines are derived from crosses with at least one or more parents with acceptable end-use quality. As soon as individual lines are identified (F_5) generation, microquality analyses are conducted and approximately 10% are discarded on the basis of poor end-use quality. In the F_6 and later generations, samples are composited from three or more locations/year, milled on a Buhler Mill, and baked using 100 g of flour per loaf. Though genotype-by-environmental interactions are large for end-use quality traits, composite samples are satisfactory for determining the end-use quality when repeated over time. By using phenotypic selection, the program has released cultivars with acceptable quality involving known 'poor' quality genes and chromosomes, such as high-molecular weight (HMW) glutenin subunits 2+12 (Scout 66 and Lancota), 1BL.1RS (heterogeneous in Rawhide and homogeneous in Cougar), and 1AL.1RS (heterogeneous in Nekota and Niobrara). Phenotypic selection is preferred to genotypic selection.

Introduction

The cooperative University of Nebraska-USDA ARS wheat breeding effort has four criteria that every new cultivar release must meet, namely, improved agronomic performance relative to existing cultivars, exceptional winterhardiness, resistance to *Puccinia graminis* (the causal agent of stem rust), and acceptable end-use quality. This paper will concentrate on breeding for acceptable end-use quality with the understanding that the end-use quality determined by the flour's ability to make leavened bread products. Acceptable end-use quality, rather than enhanced end-use quality is the goal because currently there are insufficient economic incentives to develop only cultivars with enhanced end use quality. Also, consistency of end-use quality is more desired by the baking industry than is altered quality.

The breeding program and selection for end-use quality

The breeding program is outlined in Table 1.

In general, the breeding program uses single and three-way crosses. At least one parent of the single cross will be a Nebraska-developed line and usually two parents of the three-way cross will be from Nebraska or Kansas and selected for good end-use quality. All lines which are unadapted or have different end-use quality characteristics (e.g. soft wheats or poor quality hard wheats) are used as parents in three way crosses. A single cross with an unadapted or poor quality line, generally does not have progeny with sufficient adaptation or quality genes at a high enough frequency for successful selection. This narrowness of parents may limit our germplasm, but Nebraska has very stringent winterhardiness requirements and usually populations from single crosses with an unadapted parent (which also often have unacceptable end-use quality characteristics for our purposes) are winterkilled.

Table 1. How a new wheat cultivar is developed

Year 1: Make between 600 to 900 crosses at Lincoln in the greenhouse to produce F_1 seed.

Year 2: Grow the F_1 seed in the Lincoln greenhouses to avoid losses due to winterkilling if the seed was grown in the field. Harvest F_2 seed.

Year 3: Plant F_2 seed in bulk populations at Mead, NE (50 km north of Lincoln). Mead is the most severe winter site. Infect plants with stem rust. Hence wintertender and stem rust susceptible plants will be severely injured or killed.

Year 4: Plant F_3 seed in bulk populations at Mead, NE. Infect plants with stem rust. Hence wintertender and stem rust susceptible plants will be severely injured or killed. Send 30 populations to the USDA-ARS (Manhattan, KS) to select Hessian fly resistant material. Select 45,000 heads from F_3 bulks.

Year 5: Plant 45,000 F_4 head rows at Mead, NE. Infect plants with stem rust. Wintertender and stem rust susceptible plants will be severely injured or killed. On the basis of plant type and disease resistance, harvest 1,800 head rows. Evaluate harvested seed and select 1,500 lines for advancement.

Year 6: Plant 1,500 observation F_5 plots at Lincoln, NE. All lines are screened in the greenhouse for stem rust. On the basis of plant type, yield, and disease resistance, harvest 400–450 plots. Evaluate harvested seed using microquality analyses (flour protein and Mixograph) in the Nebraska Wheat Quality Laboratory and select 285 lines for advancement that have acceptable end-use quality.

Year 7: Plant 285 F_6 lines and 15 checks (total of 300 lines) in single replication trials at seven Nebraska locations (Mead, Lincoln, Clay Center, McCook, Grant, Sidney, and Alliance). Send seed to USDA-ARS Cereal Rust Laboratory (St. Paul, MN) for stem rust testing and to Kansas State University for soilborne wheat mosaic virus testing. On the basis of plant type, yield, disease resistance, and end-use quality select about 56 lines for advancement. Evaluate harvested seed using a full milling and baking procedure at the Nebraska Wheat Quality Laboratory.

Year 8: Plant 56 F_7 lines and 4 checks (total of 60 lines) in replicated and observation trials at eight Nebraska locations (Mead, Lincoln, Clay Center, North Platte, McCook, Grant, Sidney, and Alliance). Send seed to USDA-ARS Cereal Rust Laboratory for stem rust testing and USDA-ARS for Hessian fly testing. On the basis of plant type, yield, end-use quality, and disease resistance, select about 25 lines for advancement. Evaluate harvested seed using a full milling and baking procedure at the Nebraska Wheat Quality Laboratory.

Year 9: Plant 60 F_8 to F_{12} lines in replicated and observation trials at eight Nebraska locations (Mead, Lincoln, Clay Center, North Platte, McCook, Grant, Sidney, and Alliance). The 60 lines include 10 to 15 check lines, 25 lines retained from the previous year's trials and the 25 newly advanced lines. Send seed to USDA-ARS Cereal Rust Laboratory for stem rust testing and USDA-ARS for Hessian fly testing. Test for wheat streak mosaic virus tolerance. On the basis of plant type, yield, end-use quality, and disease resistance, select 35–40 lines (including checks) for retention. Evaluate harvested seed using a full milling and baking procedure at the Nebraska Wheat Quality Laboratory. Increase seed of 10 lines for advancement to regional nurseries.

Year 10: Plant 60 F_8 to F_{12} lines in replicated and observation trials at eight Nebraska locations (Mead, Lincoln, Clay Center, North Platte, McCook, Grant, Sidney, and Alliance). The 60 lines include 10 to 15 check lines, 25 lines retained from the previous year's trials and the 25 newly advanced lines. Send seed to USDA-ARS Cereal Rust Laboratory for stem rust testing and USDA-ARS for Hessian fly testing. Test for wheat streak mosaic virus tolerance. On the basis of plant type, yield, end-use quality, and disease resistance, select 35–40 lines (including checks) for retention. Evaluate harvested seed using a full milling and baking procedure at the Nebraska Wheat Quality Laboratory. Submit 8–10 lines to regional nurseries and receive regional data. Retain 6 lines for second year testing in regional nurseries. Submit 4 lines to Nebraska state cultivar testing.

Year 11: Plant 60 F_8 to F_{12} lines in replicated and observation trials at eight Nebraska locations (Mead, Lincoln, Clay Center, North Platte, McCook, Grant, Sidney, and Alliance). The 60 lines include 10 to 15 check lines, 25 lines retained from the previous year's trials and the 25 newly advanced lines. Send seed to USDA-ARS Cereal Rust Laboratory for stem rust testing and USDA-ARS for Hessian fly testing. Test for wheat streak mosaic virus tolerance. On the basis of plant type, yield, end-use quality, and disease resistance, select 35–40 lines (including checks) for retention. Evaluate harvested seed using a full milling and baking procedure at the Nebraska Wheat Quality Laboratory. Submit 8–10 to regional nurseries and receive regional data. Retain 6 lines for second year testing in regional nurseries. Submit 4 lines to state cultivar testing. Begin Foundation Seed production of advanced lines.

Table 1. Continued

Year 12: Plant 60 F_8 to F_{12} lines in replicated and observation trials at eight Nebraska locations (Mead, Lincoln, Clay Center, North Platte, McCook, Grant, Sidney, and Alliance). The 60 lines include 10 to 15 check lines, 25 lines retained from the previous year's trials and the 25 newly advanced lines. Send seed to USDA-ARS Cereal Rust Laboratory for stem rust testing and USDA-ARS for Hessian fly testing. Test for wheat streak mosaic virus tolerance. On the basis of plant type, yield, end-use quality, and disease resistance, select 35–40 lines (including checks) for retention. Evaluate harvested seed using a full milling and baking procedure at the Nebraska Wheat Quality Laboratory. Submit 8–10 to regional nurseries and receive regional data. Retain 6 lines for second year testing in regional nurseries. Submit 4 lines to state cultivar testing. Continue Foundation Seed increase of advanced lines. If performance warrants release, release one line as a new cultivar.

A breeding program is a continuum; hence lines are constantly added and dropped from consideration. Of the 25 lines advanced in year 8, only 10–15 will be retained in year 9, 5–10 will be retained in year 10, 5 will be retained in year 11, and one or two in year 12. On average, over 100,000 lines will be looked at to find a cultivar. Over 12,000 yield plots will be harvested each year. A cultivar will be tested in over 100 location-years before we know enough to release it. It takes a minimum of 12 years to create a new wheat cultivar.

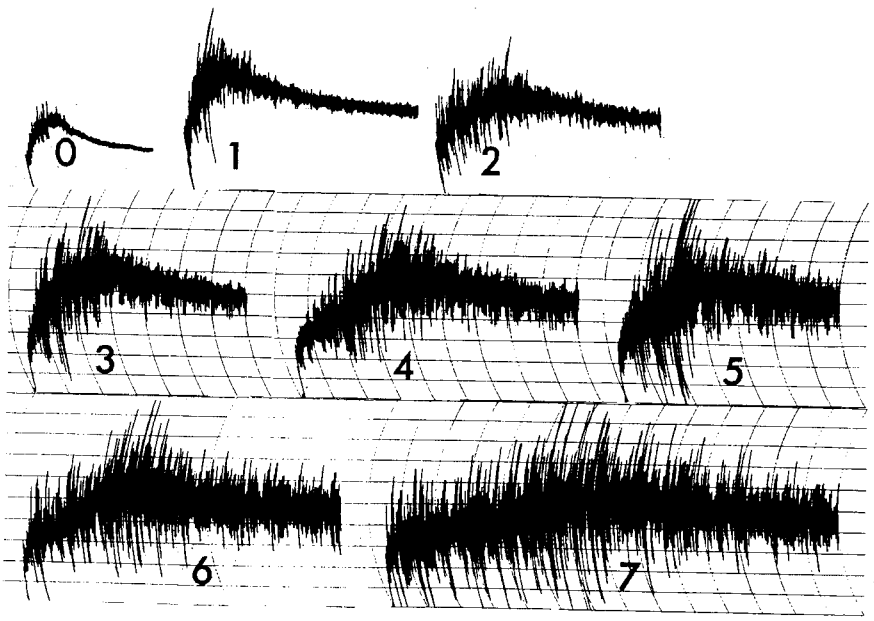

Figure 1. Mixograph curves used for scoring Mixograph tolerance (0 is considered weak and 7 is considered strong).

End-use quality assays begin in year 6. Due to a limited amount of seed (average plot yields are between 400 and 800 g/plot which is the equivalent of 2700 to 5400 kg/h), the initial tests are microquality analyses. Our microquality analyses use a 35-g grain sample from each of the selected lines is tempered to a moisture basis of 152 g H_2O kg^{-1} grain and milled in a Quadrumat Jr. Laboratory mill (C. W. Brabender Instruments Inc., OHG, Germany). Flour is separated from bran using a Strand shaker (Strand Shaker Co. Minneapolis, MN) at 225 rpm for 90 s with a standard sieve no. 70. Mixograph analyses are obtained using a National Manufacturing Mixograph (Lincoln, NE) with a 10-g sample and constant water absorption of 620 g H_2O kg^{-1} grain. Mixograph mixing time (hereafter referred to as mixing time) is recorded as the time in minutes to maximum curve height. Mixograph mixing tolerance (hereafter referred to as mixing tolerance) is rated based on comparisons against standard curves in the Nebraska Wheat Quality Laboratory using a scale from 0 to 7 with higher scores indicating greater tolerance of dough to overmixing (Method 54–40; Am. Assoc. of Cereal Chemists, 1983). Flour protein content, expressed on a 140 g H_2O kg^{-1} flour moisture basis, was determined by Udy dye binding (Udy dye Method 46–14A; Am. Assoc. of Cereal Chemists, 1983), randomly calibrated with standard Kjeldahl procedures (Method 46–10; Am. Assoc. of

Cereal Chemists, 1983) or more recently with combustion techniques (Method 46–30; Am. Assoc. of Cereal Chemists, 1995; LECO Manufacturing Equipment, St. Joseph, MI, USA). Our microquality goal is to have a mixing time of greater than 3 minutes (preferably greater than 4) and a mixing tolerance of greater than 3 (preferably greater than 4, Figure 1), with a protein level of 12% in the whole grain. The protein measurement is used to eliminate low protein lines and as a 'covariate' to help explain our mixing times and tolerances. Occasionally due to over fertilization or environmental conditions, we have elevated protein levels at Lincoln which often enhance our microquality measurements. Under these conditions, selection must be more stringent.

The selection on mixing time and tolerance is for stronger wheats because drought and heat stress during grain filling tend to reduce mixing time and tolerance (Graybosch et al., 1995), dough strength (Blumenthal et al., 1991; Blumenthal et al., 1993), and baking quality (Peterson et al., 1998). The goal is to develop strong wheats that under adverse conditions will retain acceptable end-use quality, rather than develop medium quality wheat which under adverse conditions will have unacceptable end-use quality. Hence the end-use quality of these strong wheats will be consistently acceptable. All lines are given an overall quality score of 1 (exceptional), 2 (acceptable), 3 (questionable), or 9 (unacceptable, Table 2). Normally about 10% of the selected lines are unacceptable. This figure varies and was higher when we were working with known lower quality parents (such as Siouxland (1BL.1RS) and TAM107 (1AL.1RS)).

As can be seen in Table 2, the selection intensity tends to be similar in the exceptional, acceptable, and questionable end-use quality classes. However, there is a bias to select more stringently for higher yield, as the quality becomes lower.

The microquality tests are completed in roughly 30 days (in August, after harvest and before planting). The key point of breeding for end-use quality is to eliminate poor quality lines quickly. One question that may be asked is why to do we select lines with questionable quality? We select them for two reasons. First, though the quality may be questionable by Nebraska standards, the quality is closer to our end-use quality targets than many lines we use as parents. Hence these questionable quality lines are part of our parent building program. Secondly, it is well documented that genotype-by-environment interactions are important for most wheat quality traits (Graybosch et al., 1996), so it is possible that a questionable quality line may have better quality when grown in different environments (years or locations) in the state.

Similarly, the selection intensity of exceptional and acceptable quality lines indicates that lines with lower agronomic performance are evaluated for end-use quality. If the percentage of lines that would be discarded could be accurately predicted before the microquality assay were run, fewer lines may be submitted for end-use quality assays. Also, as there is large genotype-by-environment interactions for agronomic performance, some lines which perform poorly at Lincoln (an area where semi-dwarf wheats are favored) may have attributes which will allow them to perform well elsewhere in the state (where drought requires long coleoptile length and tall plant height).

We are ruthless in discarding the unacceptable quality lines because our past experience has indicated that they are rarely or never acceptable under any environmental conditions. They could be used as parents; however, these lines have few, if any, advantages when compared to our acceptable or exceptional end-use quality lines. Hence there is no need to retain or advance unacceptable quality lines. In addition, eliminating unacceptable quality lines quickly saves resources and does not allow the developer to become attached to the line. Before we went to the 30-day completion of the microquality analyses, between 10 and 20% of the 285 lines selected for advancement were discovered before harvest to have unacceptable quality, thus were a waste of resources. An interesting question that wheat breeders often debate is whether or not breeding for end-use quality slows genetic gain or progress. Our opinion is that breeding for end-use quality does not slow genetic gain or progress, but rather increases the cost of the genetic gain. Basically more lines are harvested for possible advancement with the understanding that 10% or more will be eliminated due to unacceptable quality. The program becomes larger for two generations (headrows and observation nursery), but after that returns to the size that we would normally maintain.

The 'full' milling and baking evaluations begin in the F_7. The lines are planted in September and their evaluations are completed before March of the following year. In this generation, approximately 2500 g or more grain composited from three or more testing locations are milled on a Buhler Laboratory mill (Buhler, Inc., Minneapolis, MN) and 100 g pup loaves are baked and evaluated (Peterson et al., 1998). In this procedure, two 100 g loaves are baked at two oxid-

Table 2. Observation lines harvested at Lincoln, NE in 1998–1999 and evaluated for possible statewide testing in 1999–2000

Quality class	No. of lines	No. of lines advanced	Selection intensity	Mean grain yield (kg/h)	Flowering days after April 30	Plant Height (cm)
Exceptional	129	83	0.64	3730	22.4	109
Acceptable	235	167	0.71	3780	22.3	109
Questionable	42	30	0.71	3970	21.9	112
Poor	36	0	0.00			
Total	442	280	0.63			

ation levels and the final bake (again involving two 100 g loaves) uses the estimated best oxidation level. The baking ingredients are considered 'lean' so that the bake test magnifies the differences in the end-use quality of the flour and does not measure the quality of the other ingredients. Usually fewer than 10% of the advanced lines are considered questionable. These lines were not identified in our preliminary screen for a number of reasons. The Quadramat junior mill does not differentiate poor milling from good milling lines well. Therefore, one to two lines are too soft for milling on the Buhler Laboratory mill or larger mills. A few lines perform poorly at the lower protein levels commonly found in western Nebraska. Finally, a few lines with adequate protein and acceptable mixing time and mixing tolerance scores simply do not make a good loaf of bread.

Milling and baking evaluations on composite samples continue in each of the later generations. The quality analyses are only done on those lines which have acceptable agronomic performance and are advanced to the next generation. The quality analyses are often more costly and labor intensive than the field evaluations, so the goal is to keep the number of quality analyses to a minimum. Usually less than one experimental line is dropped from further consideration per generation due to questionable end-use quality. While these additional end-use quality tests may not be considered necessary because so few experimental lines are identified as being unacceptable, they are valuable to estimate the average or true end-use quality of the experimental lines. Usually 6 years of quality evaluations are completed on composite samples before a line is recommended for commercial release.

With the addition of white wheat cultivar development to our program, additional tests (e.g. polyphenol oxidase, and noodle color and quality) are being added to the end-use quality assays. In addition, a few tests, such as falling number, are being done on samples from each testing site. This sampling from individual sites is mainly due to the need to develop risk-management information on what level of sprouting can be expected at various sites in Nebraska where white wheat may or may not be produced.

Phenotypic vs. genotypic breeding for end-use quality

As previously discussed, the basis for selection for end-use quality is predominantly one of consistent and repetitive testing (phenotypic selection). An alternative strategy would be to use the considerable information available on the genetics of end-use quality to eliminate lines with poor potential for acceptable end-use quality (genotypic selection) and then do the milling and baking tests. We have largely avoided the genotypic selection strategy because many of the lines with poor end-use quality genotypes can be quickly eliminated using our microquality assays and those that are not eliminated with the microquality or larger end-use quality assays, may have acceptable end-use quality. Some of these lines have been released and have become highly successful cultivars. For example, Scout 66 which was one of the most widely grown and well received cultivars for end-use quality has the HMW 2 + 12 glutenin subunits which are generally considered deleterious for end-use quality. Though Siouxland (1BL.1RS) was marginally acceptable for end-use quality, Rawhide (heterogeneous for 1B and 1BL.1RS) was very acceptable for end-use quality and the 1BL.1RS genotypes within Rawhide actually were superior to many 1B wheats (Moreno-Sevilla et al., 1995). Cougar, a recent release, is 1BL.1RS wheat with acceptable end-use quality and an unreleased 1BL.1RS sister line, NE93405, actually had very good

end-use quality (data not shown). Similarly, cultivars heterogeneous for 1AL.1RS and 1A (Nekota and Niobrara) have acceptable end-use quality (Espitia-Rangel et al., 1999) and have been widely grown. Nekota has been quite popular in South Dakota and Niobrara is the second most widely grown wheat in Nebraska (1999 cultivar survey). These cultivars are too valuable too be discarded before their true worth is known.

References

AACC, 1983. Approved Methods of the AACC. AACC, St Paul, MN.

AACC, 1995. Approved Methods of the AACC. AACC, St Paul, MN.

Blumenthal, C.S., E.W.R. Barlow & C.W. Wrigley, 1993. Growth environment and wheat quality: the effect of heat stress on dough properties and gluten proteins. J Cereal Sci 18: 3–21.

Blumenthal, C.S., I.L. Batey, F. Bekes, W.W. Wrigley & E.W.R. Barlow, 1991. Seasonal changes in wheat-grain quality associated with high temperatures during grain filling. Aust J Agric Res 42: 21–30.

Espitia-Rangel, E., P.S. Baenziger, D.R. Shelton, R.A. Graybosch, B. Moreno-Sevilla & C.J. Peterson, 1999. End-use Quality Performance and Stability of 1A vs. 1AL.1RS Genotypes Derived from Winter Wheat 'Nekota'. Crop Sci 39: 649–654.

Graybosch, R.A., C.J. Peterson, D.R. Shelton & P.S. Baenziger, 1996. Genotypic and environmental modification of wheat flour protein composition in relation to end-use quality. Crop Sci 36: 296–300.

Graybosch, R.A., C.J. Peterson, P.S. Baenziger & D.R. Shelton, 1995. Environmental modification of hard red winter wheat flour protein composition. J Cereal Sci 22: 45–51.

Moreno-Sevilla, B., P.S. Baenziger, D.R. Shelton, R.A. Graybosch & C.J. Peterson, 1995. Agronomic performance and end-use quality of 1B vs. 1BL/1RS genotypes derived from winter wheat 'Rawhide'. Crop Sci 35: 1607–1612.

Peterson, C.J., R.A. Graybosch, D.R. Shelton & P.S. Baenziger, 1998. Baking quality of hard winter wheat: response of cultivars to environment in the Great Plains. Euphytica 100: 157–162.